

PAb hTLR1

Polyclonal antibody to human TLR1

Catalog # pab-hstlr1

For research use only, not for diagnostic or therapeutic use

Version # 07E15-MT

PRODUCT INFORMATION

Content

• 200 µg polyclonal anti-hTLR1 antibody (PAb-hTLR1), provided sterile, azide-free and lyophilized.

Isotype: Rat IgG

Formulation: H₂O with 250 U/ml Pen and 250 µg/ml Strep

Antibody resuspension

Add 1 ml of sterile PBS to obtain a concentration of 0.2 mg/ml.

Storage

- Product is shipped at room temperature. Lyophilized PAb-hTLR1 should be stored at -20°C. Product is stable for 6 months.
- Resuspended PAb-hTLR1 should be stored at 4°C for 1 month or -20°C for 3 months.

Description

PAb hTLR1 is a polyclonal antibody specific for human Toll-like receptor 1 (TLR1, CD281). PAb hTLR1 was generated by DNA vaccination. Wistar rats received four hydrodynamic injections of pVAC-hTLR1, a plasmid expressing the extracellular region of human TLR1. The sera were harvested and the IgG fraction purified by Protein G affinity chromatography.

BACKGROUND

TLR1 is predominantly expressed in the spleen and peripheral blood cells. No direct ligands have been identified so far for TLR1, and its function remains unclear. TLR1 seems to act as a coreceptor for TLR2. TLR1 and TLR2 form heterodimeric complexes on the cell surface and in the cytosol¹. TLR1 and TLR2 were shown to cooperate in recognizing *Borrelia burgdorferi* outer-surface protein A lipoprotein OspA². They also interact to recognize the 19-kD mycobacterial lipopeptide and several synthetic triacylated lipopeptides³, but not diacylated lipopeptides. This suggests that TLR1 is able to discriminate among lipoproteins by recognizing the lipid configuration⁴.

References

1. Sandor F. *et al.*, 2003. Importance of extra- and intracellular domains of TLR1 and TLR2 in NFκB signaling. *J Cell Biol.* 2003 Sep 15;162(6):1099-110.
2. Alexopoulou L. *et al.*, 2002. Hyporesponsiveness to vaccination with *Borrelia burgdorferi* OspA in humans and in TLR1- and TLR2-deficient mice. *Nat Med.* 8(8):878-84.
3. Takeuchi O. *et al.*, 2002. Cutting edge: role of toll-like receptor 1 in mediating immune response to microbial lipoproteins. *J Immunol.* 169(1):10-4.
4. Takeuchi O. *et al.*, 2001. Discrimination of bacterial lipoproteins by Toll-like receptor 6. *Int Immunol.* 13(7):933-40.
5. Schindler U. & Baichwal VR., 1994. Three NF-κB binding sites in the human E-selectin gene required for maximal tumor necrosis factor alpha-induced expression. *Mol Cell Biol.* 14(9):5820-5831.

APPLICATIONS

PAb hTLR1 can be used for neutralization of TLR1, it blocks the cellular activation of TLR1 induced by TLR1/TLR2 agonists, such as Pam3CSK4. Other applications have not been tested.

Neutralization Protocol

Neutralization experiments were performed in THP1 cells, a human monocytic cell line that naturally expresses TLR1 and TLR2, and HEK293 cells (which express TLR1 endogenously) transfected to stably express human TLR2. These cells were further transfected with pNiFty-SEAP, a plasmid that expresses a secreted embryonic alkaline phosphatase (SEAP) gene under the control of an NF-κB-inducible ELAM-1 (E-selectin) promoter⁴. The amount of SEAP secreted in the supernatant can be readily detected when using QUANTI-Blue™, a SEAP detection medium. QUANTI-Blue™ will turn blue following TLR stimulation but remain pink if neutralization occurs.

Procedure for HEK293/TLR2-SEAP cells

- 1- Prepare a 1/10 PAb-hTLR1 dilution (20 µg/ml) using culture medium with heat inactivated FBS.
- Note: Some lots of FBS contain endogenous alkaline phosphatase that can interfere with SEAP.*
- 2- Prepare a cell suspension at 250,000 cells/ml.
- 3- Add 100 µl of cell suspension per well of a 96-well plate.
- 4- Add 100 µl of PAb-hTLR1 dilution (5 µg/ml final).
- 5- Incubate 10 min at 37°C.
- 6- Add 5 ng/ml of Pam3CSK4.
- 7- Incubate overnight at 37°C
- 8- Add 50 µl supernatant to 150 µl QUANTI-Blue™ in a 96-well plate.
- 9- Incubate 15-30 min at 37°C
- 10- Assess SEAP levels with the naked eye or spectrophotometrically by reading the OD at 655 nm.

RELATED PRODUCTS

Product	Catalog Code
MAb hTLR1 (monoclonal)	mab-htlr1
PAb hTLR2 (polyclonal)	pab-htlr2
293/hTLR2	293-htlr2
pUNO-hTLR1 (human gene)	puno-htlr1
pUNO-hTLR2 (human gene)	puno-htlr2
pNiFty-SEAP	pnifty-seap
QUANTI-Blue™	rep-qb-1
Pam3CSK4	tlr1-pms

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